510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE

A. 510(k) Number:

k100101

B. Purpose for Submission:

New device

C. Measurand:

Galactose-1-phosphate uridyl transferase (GALT) activity

D. Type of Test:

Quantitative, time-resolved fluoroimmunoassay

E. Applicant:

PerkinElmer, Inc.

F. Proprietary and Established Names:

GSP Neonatal GALT kit

G. Regulatory Information:

1. Regulation section:

   21 CFR 862.1315 Galactose-1-phosphate uridyl transferase test system

2. Classification:

   II

3. Product code:

   KQP

4. Panel:

   Chemistry

H. Intended Use:

1. Intended use(s):

   The GSP Neonatal GALT kit is intended for the quantitative determination of galactose-1-phosphate uridyl transferase (GALT) activity in blood specimens dried on filter paper as an aid in screening newborns for classical galactosemia caused by GALT deficiency using the GSP instrument.

2. Indication(s) for use:

   See intended use above.

3. Special conditions for use statement(s):

   For prescription use only.

   As with any other *in vitro* screening test, the data obtained using the GSP Neonatal GALT kit should be used as an aid to other medically established procedures and results interpreted in conjunction with other clinical data available to the clinician. A diagnostic procedure should be used to confirm a diagnosis of classical galactosemia.

4. Special instrument requirements:

   Only for use on the GSP Instrument

I. Device Description:

The GSP Neonatal GALT kit consists of the following reagents (to perform 1152 assays):

- **Neonatal GALT calibrators:** prepared from sheep blood with GALT, phosphoglucomutase, glucose-6-phosphate dehydrogenase, DTT, and ProClin 300 as preservative. The hemoglobin concentration is approximately 170 g/L prior to dispensing onto filter paper cassettes.

  The six calibrators contain GALT activities of approximately 1, 3, 6, 9, 15 and 25 U/dL.

- **Neonatal GALT controls:** prepared from human and sheep blood with preservative. The hemoglobin concentration is approximately 170 g/L prior to dispensing onto filter paper cassettes.

  The two controls contain approximate GALT activities of 4 and 13 U/dL.

- **GALT Substrate Reagent** - ß-nicotinamide adenine dinucleotide phosphate, uridine 5'-diphosphoglucose, galactose-1-phosphate, and DTT.

- **GALT Assay Buffer -** a ready-for-use buffer containing magnesium sulfate, EDTA, tris (hydroxymethyl) aminomethane, Triton X-100, and preservative.

This kit contains calibrators manufactured from sheep blood components and controls manufactured from sheep and human blood components. The sheep are from a closed herd located in the USA. The human blood has been tested using FDA approved methods or equivalent and found to be negative for hepatitis B surface antigen, anti-hepatitis C and anti-HIV 1 and 2 antibodies

J. Substantial Equivalence Information:

1. Predicate device name(s):

   PerkinElmer Neonatal GALT (formerly Isolab Galactose-1-phosphate uridyl tranferase test kit)

2. Predicate K number(s):

   k950803

3. Comparison with predicate:

Similarities		
Characteristic	**Predicate device (k950803)**	**Proposed Device (k950803)**
Indications for use	An aid in screening newborns for classical galactosemia caused by GALT deficiency	Same
Test Mode	Batch mode	Same
Detection Technology	Prompt fluorescence	Same
Sample Type	Dried blood spots.	Same
Number of Calibrators	Six	Same
Calibrator matrix	Sheep blood with GALT, phosphoglucomutase, glucose-6-phosphate dehydrogenase, dithiothreitol and ProClin 300.	Same
Number of Controls	Two	Same
Substrate Reagent	Beta-nicotinamide adenine dinucleotide phosphate,uridine 5'-diphosphoglucose, galactose-1-phosphate, and dithiothreitol	Same

Differences		
Characteristics	**Predicate Device (k950803)**	**Proposed Device**
Analytical method	Semi-quantitative	Quantitative
Instrument Platform	1420 Victor D series fluorometer	GSP instrument
Calibrator and Control Format	Filter paper sheets (Whatman no. 903)	Filter paper cassettes (Whatman no.903)

Calibrator Concentrations	A 1.8 U/g Hb B 5 U/g Hb C 8 U/g Hb D 11 U/g Hb E 14 U/g Hb F 18 U/g Hb	A 1 U/dL B 3 U/ dL C 6 U/ dL D 9 U/ dL E 15 U/ dL F 25 U/ dL
Control Matrix	Sheep blood with GALT, phosphoglucomutase, glucose-6-phosphate dehydrogenase and dithiothreitol with ProClin 300 as preservative.	Human and sheep blood with ProClin 300 as preservative
Control Concentrations	Approx. values: Normal 12.7 U/g Hb Abnormal 2.1 U/g Hb	Approx. values: Low 4 U/dL High 13 U/dL
Reconstitution Buffer	Ready-for-use buffer contains magnesium sulphate, ethylenediaminetetraacetic acid, tri aminomethane, and preservative.	Ready-for-use buffer contains magnesium sulfate, ethylenediaminetetraacetic acid, tris aminomethane, Triton X-100, and preservative.
MicroPlates	Black uncoated	Clear uncoated, sold separately
Calculation	The system incorporates programs for data reduction, and the results obtained as printouts of calibration curves, unknown activities etc.	GSP Workstation software, X-axis LIN, Y-axis LIN; fitting algorithm linear regression
Incubation Detail	3 hours, 37˚C and 60min, RT	20 min + 2 hours, 37°C
Testing Integrity Controls	Not available	Floating Disk Control – detects floating sample disks in the wells before measuring GALT activity Elution Control - detects missing sample disks in the wells after measuring GALT activity

K. Standard/Guidance Document Referenced (if applicable):

- CLSI Guideline EP5-A2: *Evaluation of Precision Performance of Quantitative Measurement Methods*
- CLSI Guideline EP 17-A: *Protocols for Determination of Limits of Detection and Limits of Quantitation*

- CLSI Guideline EP6-A: *Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach*
- CLSI Guideline EP9-A2*: Method Comparison and Bias Estimation Using Patient Samples*
- CLSI Protocol EP7-A2: *Interference Testing in Clinical Chemistry*

L. Test Principle:

The GSP™ Neonatal GALT assay is an adaptation of the enzymatic assay of Beutler and Baluda. A summary of the reaction sequence of the GALT assay is shown below. The enzymes shown above the arrows are found in the sample itself. All others are either products of the previous reaction or found in one of the reagents. The GSP Neonatal GALT assay uses Prompt Fluorescence technology.

$$\text{GAL-1-P + UDPG} \xrightarrow{\text{GALT}} \text{G-1-P + UDP-GAL}$$

$$\text{G-1-P} \xrightarrow{\text{PGM}} \text{G-6-P}$$

$$\text{G-6-P + NADP} \xrightarrow{\text{G-6-PD}} \text{6-PGA + NADPH*}$$

$$\text{6-PGA + NADP} \xrightarrow{\text{6-PGD}} \text{R-5-P + NADPH*}$$

GAL-1-P= Galactose-1-phosphate
UDPG = Uridine 5'-diphosphoglucose
GALT = Galactose-1-phosphate uridyl transferase
G-1-P = Glucose-1-phosphate
UDP-GAL = Uridine diphosphogalactose
PGM = Phosphoglucomutase
G-6-P = Glucose-6-phosphate
NADP = Nicotinamide adenine dinucleotide phosphate
G-6-PD = Glucose-6-phosphate dehydrogenase
6-PGA = 6-phosphogluconate
NADPH = Nicotinamide adenine dinucleotide phosphate (reduced)
* = Fluorescent
6-PGD = 6-PGA dehydrogenase
R-5-P = Ribulose-5-phosphate

The fluorescence is measured using an excitation wavelength of 355 nm and an emission wavelength of 460 nm.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

   a. Precision/Reproducibility:

   Whole blood samples with clinically relevant GALT activities were used to prepare dried blood spot samples. The samples (S1-S8) were chosen to cover the measuring range of the GSP Neonatal GALT kit, which is from 2.5 to 25.0 U/dL. Samples 1-4 (S1-S4) were chosen to cover the medical decision making level.

   The GALT activities of the series of dried blood spot samples were measured by three laboratorians with three GSP Neonatal GALT kit lots in 27 runs over 25 operating days using three GSP instruments (9 runs per instrument). Each sample was measured in quadruplicates.

   In this study one run equals a run of two plates tested 24 hours apart on the same instrument with stored calibration. Within a run only one reagent lot was used in order to assess the variation between lots. Separate estimations for the precision of GSP Neonatal GALT assays were calculated with two options: a full calibration curve in duplicate for each plate and a full calibration curve for every batch of two plates.

Sample	n	Mean GALT activity (U/dL)	Within run variation		Within lot variation		Total variation	
			SD	CV%	SD	CV%	SD	CV%
S1	209*	2.5	0.3	10.3	0.4	14.3	0.4	15.9
S2	216	3.1	0.3	8.6	0.4	12.6	0.4	13.1
S3	206*	3.9	0.2	6.1	0.3	8.3	0.3	8.6
S4	216	5.3	0.3	5.7	0.5	9	0.5	9.2
S5	216	7.1	0.3	3.8	0.5	7.4	0.5	7.6
S6	216	13.1	0.9	6.7	1.2	8.9	1.2	9.1
S7	216	18.3	0.6	3.1	0.9	5.1	1.0	5.4
S8	216	22.5	0.7	3.0	1.1	4.9	1.2	5.2

Precision data using one calibration curve valid for 24 h.

Sample	n	Mean GALT activity (U/dL)	Within run variation		Within lot variation		Total variation	
			SD	CV%	SD	CV%	SD	CV%
S1	209*	2.4	0.3	12.8	0.3	14.3	0.4	15.7
S2	216	3	0.4	12.2	0.4	13.3	0.4	13.6
S3	206*	3.9	0.3	7.6	0.3	8.8	0.3	9
S4	216	5.3	0.4	7.9	0.5	9.6	0.5	9.8
S5	216	7	0.4	5.7	0.6	7.9	0.6	8.3
S6	216	13	1	7.4	1.2	9	1.2	9.5
S7	216	18.2	0.7	3.8	1.0	5.5	1.1	6.0
S8	216	22.4	0.8	3.4	1.2	5.3	1.3	5.8

* The sponsor stated that occasionally, a sample punched from a DBS does not sink to the bottom of the plate well, but remains floating on the liquid surface. The GALT activity measured from a well with a floating sample disk is not reliable and during routine use the instrument will flag the sample. The samples having floating disks were not rerun.

b. Linearity/assay reportable range:

The linearity study protocol and analysis for the GSP Neonatal GALT kit was performed in accordance with CLSI document EP6-A: *Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach, Approved Guideline*.

The claimed measuring range for this device is 2.50 U/dL to 25 U/dL.

A high activity sample was prepared by diluting sheep erythrocytes to Hb 17 g/L following addition of GALT, PGM and G6PD enzymes. The low activity sample contained sheep red blood cells without any added enzymes. 18 samples were prepared that spanned the claimed measuring range by mixing the high activity and low activity samples.

The series of samples were used to prepare dried blood spot samples by dispensing the prepared samples onto filter paper and dried overnight.
The dried blood spot samples were tested in a single run in random order on one day with one GSP Neonatal GALT kit lot. The samples were analyzed in 10 replicates.

A polynomial evaluation of linearity was used for the data analysis. The assumption of constant variance across all levels is not fulfilled in the GSP Neonatal GALT kit. Rather, the variance is proportional across different measurement levels. Therefore, weighted regression models were used.

A linear regression line and second and third order polynomials were fitted to the data. The results of regression analyses were compared. The significance of the second and third order polynomials were evaluated by performing a t-

test. The third order regression has statistically significant nonlinear terms ($\beta 2$, $\beta 3$) at a 95% significance level (p-value <0.05).

The results of regression analyses were compared. The fitted models are:

Linear: $y = -0.68 + 34x$

Second order: $y = -0.73 + 35x - 1x^2$

Third order: $y = -0.21 + 28x + 22x^2 - 20x^3$

where y = GALT activity (U/dL) and x = dilution point.

The comparison of the measured and expected results are presented below:

Dilution	Measured GSP GALT (U/dL serum)	Linear model predicted GSP GALT (U/dL serum)	3rd order model predicted GSP GALT (U/dL serum)	Absolute difference between models (ng/mL serum)	Relative difference between models (%)
0.09	2.54	2.42	2.48	0.07	2.9
0.12	3.49	3.45	3.45	0.00	-0.1
0.16	4.54	4.82	4.77	-0.06	-1.2
0.2	6.28	6.2	6.13	-0.07	-1.2
0.3	9.87	9.64	9.65	0.01	0.1
0.4	13.3	13.1	13.3	0.17	1.3
0.5	16.9	16.5	16.8	0.30	1.8
0.6	20.1	20	20.2	0.28	1.4
0.7	22.8	23.4	23.4	-0.02	-0.1
0.8	26.7	26.8	26.2	-0.70	-2.6

For GALT activities over 4 U/dL, the maximum observed difference (%) between the linear and 3rd order regression models is -2.6 %. For activities ≤ 4 U/dL, the maximum observed absolute difference between the models is 0.07 U/dL.

For GSP Neonatal GALT, the method has demonstrated the claimed measuring range of the device as 2.5 – 25.0 U/dL.

c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*

Traceability: There is no international conventional reference material that can be used as the primary calibrator and no reference method that can be used to assign values. Instead the GALT activity of the kit is traceable to the in-house primary calibrators (dried blood spots). The activity of these

calibrators is based on Galactose-1-phosphate Uridyltransferase from galactose-adapted yeast, added to the calibrator blood matrix.

Value assignment: The in-house reference materials used in the calibration process of GSP Neonatal GALT kit include primary, secondary and level calibrators and controls (QA and R controls). The primary calibrators are used to monitor the level of the secondary and level calibrators.

The secondary calibrators are routinely used to establish the master curve from which a new batch (lot) of kit calibrators will be assigned values. (QA controls are used to validate the run.)

To accomplish assigning reagent lot specific values, level calibrators are measured against the reagent lot calibration curve. Based on these results, the values assigned for the kit calibrators are modified in such manner that each reagent kit lot will display the same result when the level calibrators are used as samples. (QA controls are used to validate the runs.)

Stability:

Accelerated and real-time shelf-life stability: Study protocols, preliminary data and acceptance criteria for shelf-life stability testing were provided for the Neonatal GALT kit components at the recommended storage temperature (calibrators and controls at -30 to -16°C and kit reagents at 2-8°C) and at elevated temperatures (4°C for calibrators and controls; 35°C for kit reagents) and found to be acceptable. Based on the accelerated stability data, twelve month shelf-life stability at the recommended storage temperatures is claimed for the calibrators, controls and kit reagents. Real-time studies will continue to confirm and extend the dating.

In-use and on-board stability: Study protocols, preliminary data and acceptance criteria for in-use and on-board stability were provided for the Neonatal GALT kit and found to be acceptable. Once opened, calibrators and controls can be stored for 16 days at +2 to +8 °C in the original bag (protected from light and moisture), and thereafter used in the GALT assay. Stability data supports on-board storage of reagent cassette components (reconstituted Substrate Reagent and GALT Assay buffer) up to 48 hours. Neonatal GALT kit calibrators and controls punched into microtiter wells and on-board in the plate storage module of the GSP instrument are stable up to 12 hours.

d. Detection limit:

The analysis of the limits of blank, detection and quantitation were performed in accordance with CLSI document EP17-A: *Protocols for Determination of Limits of Detection and Limits of Quantitation*.

Limit of Blank (LoB): Three lots of a blank specimen (blood from one adult donor who is GALT deficient (classical galactosemia)) which contains no GALT enzyme activity were used in determination of LoB. Dried blood spot

samples were prepared by dispensing the blank blood preparations on filter paper and dried. Repeated measurements (n = 90/sample) were carried out using three analyte free dried blood spot samples. The samples were measured with three GSP Neonatal GALT kits using three GSP instruments. Nine separate runs of three samples with ten replicates of each were performed over three operating days. The samples were assayed against a full calibration curve in each run.

The LoB for each kit lot tested was calculated separately. The largest LoB of the three kit lots is used as the estimate. LoB is estimated to be 1.6 U/dL.

Limit of Detection (LoD) and Limit of Quantitation (LoQ): To prepare the low-level samples for LoD and LoQ study, blood drawn from four apparently healthy adults (with normal GALT activity) and blood drawn from an adult with GALT deficiency were used. Five low activity samples were prepared by diluting the normal samples with the GALT deficient specimen. The low-level samples were dispensed on filter paper and dried.

For LoD repeated measurements n=1080 (n = 216/sample) were carried out using five low level samples (dried blood spot samples on filter paper). Altogether 27 runs were assayed over 25 operating days using three GSP instruments and three GSP Neonatal GALT kit lots. The samples were assayed against a full calibration curve. Two QA controls run in quadruplicate (n = 4) were included in each plate and used for run acceptance. The low level samples used to define the LoD samples were also used for determining the LoQ.

The most conservative approach was taken in reporting the result. Accordingly the largest LoD of the three kit lots is used as the estimate. Thus, LoD was calculated to be 2.5 U/dL.

In absence of a recognized reference method, a functional sensitivity study was used to define LoQ. LoQ is the lowest activity of GALT that can be measured with acceptable total variation of the assay. The specification for total variation (CV% ≤ 20 %) is fulfilled for all samples with activities equal or higher than 1.8 U/dL, therefore,

LoQ = LoD

Therefore LoQ is estimated to be 2.5 U/dL.

The measuring range is demonstrated to be from 2.5 to 25 U/dL based on the Limit of Quantitation and Linearity studies. Samples that result in values below 2.5 U/dL are reported as "<2.5 U/dL". These results are not recommended to be considered accurate, but the specimen can be considered screen positive for classical galactosemia.

Samples that result in values above 25 U/dL are reported as ">25 U/dL". These results are not recommended to be considered accurate, but the specimen can be considered screen negative for classical galactosemia.

e. Analytical specificity:

The effect of the most essential interfering substances, bilirubin (unconjugated and conjugated), lipemia, total protein, glutathione, ascorbic acid, galactose and galactose-1-phosphate were evaluated. The characterization of the susceptibility of the GSP Neonatal GALT assay to interference was performed in accordance with CLSI document EP7-A2, *Interference Testing in Clinical Chemistry; Approved Guideline.*

The test was performed using one lot of the GSP Neonatal GALT kit. Three whole blood samples with clinically relevant GALT activities were used for preparing dried blood spot samples (DBS) for the study.

The interfering substances and their highest concentrations tested are described in the table below:

Interference	**Added substance**	**Approximate concentration of High pool**
Bilirubin	Unconjugated Bilirubin	40 mg/dL
Bilirubin	Conjugated bilirubin (ditaurobilirubin)	40 mg/dL
Lipemia	Intralipid	1000 mg/dL
Total protein	BSA	4000 mg/dL
Total protein	HSA	4000 mg/dL
Glutathione	Glutathione	75 mg/dL
Ascorbic Acid	Ascorbic Acid	3 mg/dL
Galactose	Galacose	50 mg/dL
Galactose-1-phosphate	Galactose-1-phosphate	50 mg/dL

The tested substances were added to the three samples to create test sample pools with high concentration of the interferent. Control pools were prepared by adding the same amount of solvent to the test samples as was added to create the high test pools. The blood pool with high concentration of added substance and the control pool were used for preparing a dilution series in order to measure the effect of the concentration of the potential interfering factor. The five-level dose-response series was prepared. The GALT activities of the dried blood spot samples were analyzed in twelve replicates using one GSP Neonatal GALT kit lot.

Potential interference was evaluated using the paired-difference method. If after evaluating potential interferents with this method, it appeared that the added substance was shown to cause interference, the dilution series of the tested substance were analyzed and the effect of the concentration of the interfering factor was assayed. BSA, HSA, glutathione and galactose were found to interfere with the measurement of GALT, therefore the dose-response method was used to test these substances further.

Icteric (unconjugated bilirubin (≤ 40 mg/dL), conjugated bilirubin (≤ 40 mg/dL)) and lipemic (Intralipid solution, ≤ 1000 mg/dL) samples did not interfere with the assay.

Ascorbic acid (≤ 3 mg/dL) and galactose (≤ 50 mg/dL) did not interfere with the assay at tested concentrations.

Glutathione did not interfere up to concentration of 18.8, 37.5 and 56.3 mg/dL at sample GALT activities of 3, 6 and 12 U/dL, respectively. Glutathione concentrations above these levels caused a decrease of up to 63% in GALT activity.

Galactose-1-phosphate (GAL-1-P) had no effect on the low GALT activity sample (3 U/dL), while a GAL-1-P concentration of 12.5 mg/dL interfered with the result of the samples with GALT activities 6 and 12 U/dL. The measured GALT result decreased up to 37%.

Total protein (HSA) had no effect on the high (12 U/dL) GALT activity sample. HSA did not interfere up to added concentration of 3000 mg/dL blood, which is approximately two times higher than the normal endogenous concentration of normal neonates, at sample GALT activities 3 and 6 U/dL. HSA concentrations above this level caused an increase up to 30% in GALT activity

Hematocrit: The effect of hematocrit was tested by adjusting the amount of red blood cells with plasma in three whole blood samples with different GALT activities (approximately <1, 6 and 15 U/dL), and testing the samples for GALT activity according to CLSI document EP7-A, see table below.

Hematocrit % (approximate value)	Sample 1		Sample 2		Sample 3	
	U/dL	n	U/dL	n	U/dL	n
35	0.99	12	7.3	12	13.2	12
44	0.37	12	6.5	11	14.9	12
53	0.00	12	5.6	12	15.4	10
62	0.00	11	5.0	12	15.6	12
70	0.00	12	4.6	11	15.2	12

Samples with low GALT activity might get slightly elevated or lowered results from the GSP Neonatal GALT assay due to differences in the hematocrit level. GALT activity is in the red blood cells and hence the GALT activity varies based on hematocrit level.

However, hemoglobin is known to absorb part of the excitation and emission light. In samples with normal GALT activity the change in hematocrit is compensated with the hemoglobin effect. In samples with low GALT activity there is not enough GALT activity to overcome the quenching effect of hemoglobin and thus the samples with low GALT activity and low hematocrit

may result in elevated results and samples with low GALT activity and high hematocrit may result in lower results.

The differences in hematocrit level have no effect on the screening classification of samples that lack GALT activity (classical galactosemia).

f. Assay cut-off:

Not applicable.

2. Comparison studies:

a. Method comparison with predicate device:

The comparator device is the Neonatal GALT (k950803) (formerly known as Isolab Galactose-1-phosphate uridyl transferase test kit). Prospective, leftover, routine screening samples as well as retrospective archived positive samples were included in the study. The total number of acquired samples is comprised of 2146 routine screening samples collected prospectively, 33 presumptive positive or borderline retrospective samples.

The clinical diagnosis for two of the retrospective presumptive positive specimens was classical galactosemia. 32 of the retrospective presumptive positive specimens were from newborns with Duarte galactosemia and one specimen was from newborn carrier for Classical Galactosemia. One routine screening specimen was confirmed to be Duarte Galactosemia.

The specimens were analyzed during 8 operating days. Each patient specimen was analyzed in singlicate using both the GSP Neonatal GALT kit and the predicate. The retrospective positive and borderline specimens were dispersed throughout the multiple assay runs. One lot of each device was used.

0.5%, 1.0%, and 1.5% percentile cut-offs were reviewed.

Screening results based on 0.5% percentile:

Classification of confirmed classical galactosemia (N=2) and other samples (N=2177) into test result categories using 0.5% percentile cut-off determination:

GSP Neonatal GALT cut-off 5.5 U/dL	Neonatal GALT cut-off 5.1 U/g Hb	Total subjects	Confirmed Classical GALT	Screening negative for Classical GALT
+	+	39*	2	37
+	-	5	0	5
-	+	3	0	3
-	-	2132	0	2132
Total		2179	2	2177

*Includes all 33 retrospective low GALT activity screening specimens.

Classification of confirmed low GALT (N=34) and other samples (N=2145) into test result categories using 0.5% percentile cut-off determination:

GSP Neonatal GALT cut-off 5.5 U/dL	Neonatal GALT cut-off 5.1 U/g Hb	Total subjects	Confirmed Low GALT	Screening negative for Low GALT
+	+	39*	33	6
+	-	5	1	4
-	+	3	0	3
-	-	2132	0	2132
Total		2179	34	2145

*Includes all 33 retrospective low GALT activity screening specimens.

Distribution of samples into test results categories using 0.5% percentile cut-off determination: GSP Neonatal GALT vs. Neonatal GALT:

0.5% cutoff	Neonatal GALT		
GSP Neonatal GALT	Test Positive	Test Negative	Total
Test Positive	39	5	44
Test Negative	3	2132	2135
Total	42	2137	2179

Overall % agreement = (39+2132)/(2179)*100% = 99.6% (CI 99.3%-99.9%)

Positive % agreement = (39/42)*100% = 92.9% (CI 83.9%-100%)

Negative % agreement = (2132/2137)*100%= 99.8% (CI 99.5%-100%)

Screening results based on 1.0% percentile:

Classification of confirmed classical galactosemia (N=2) and other samples (N=2177) into test result categories using 1.0% percentile cut-off determination:

GSP Neonatal GALT cut-off 6.7 U/dL	Neonatal GALT cut-off 5.7 U/g Hb	Total subjects	Confirmed Classical GALT	Screening negative for Classical GALT
+	+	45*	2	43
+	-	8	0	8
-	+	8	0	8
-	-	2118	0	2118
Total		2179	2	2177

*Includes all 33 retrospective low GALT activity screening specimens.

Classification of confirmed low GALT (N=34) and other samples (N=2145) into test result categories using 1.0% percentile cut-off determination:

GSP Neonatal GALT cut-off 6.7 U/dL	Neonatal GALT cut-off 5.7 U/g Hb	Total subjects	Confirmed Low GALT	Screening negative for Low GALT
+	+	45*	33	12
+	-	8	1	7
-	+	8	0	8
-	-	2118	0	2118
Total		2179	34	2145

*Includes all 33 retrospective low GALT activity screening specimens.

Distribution of samples into test results categories using 1.0% percentile cut-off determination: GSP Neonatal GALT vs. Neonatal GALT:

1.0% cutoff	Neonatal GALT		
GSP Neonatal GALT	Test Positive	Test Negative	Total
Test Positive	45	8	53
Test Negative	8	2118	2126
Total	53	2126	2179

Overall % agreement = (45+2118)/(2179)*100% = 99.3% (CI 98.9%-99.7%)

Positive % agreement = (45/53)*100% = 84.9% (CI 74.3%-95.5%)

Negative % agreement = (2118/2126)*100%= 99.6% (CI 99.3%-99.9%)

Screening result based on 1.5% percentile:

Classification of confirmed classical galactosemia (N=2) and other samples (N=2177) into test result categories using 1.5% percentile cut-off determination:

GSP Neonatal GALT cut-off 7.5 U/dL	Neonatal GALT cut-off 6.1 U/g Hb	Total subjects	Confirmed Classical GALT	Screening negative for Classical GALT
+	+	51*	2	49
+	-	14	0	14
-	+	10	0	10
-	-	2104	0	2104
Total		2179	2	2177

*Includes all 33 retrospective low GALT activity screening specimens.

Classification of confirmed low GALT (N=34) and other samples (N=2145) into test result categories using 1.5% percentile cut-off determination

GSP Neonatal GALT cut-off 7.5 U/dL	Neonatal GALT cut-off 6.1 U/g Hb	Total subjects	Confirmed Low GALT	Screening negative for Low GALT
+	+	51*	33	18
+	-	14	1	13
-	+	10	0	10
-	-	2104	0	2104
Total		2179	34	2145

*Includes all 33 retrospective low GALT activity screening specimens.

Distribution of samples into test results categories using 1.5% percentile cut-off determination: GSP Neonatal GALT vs. Neonatal GALT

1.5% cutoff	Neonatal GALT		
GSP Neonatal GALT	Test Positive	Test Negative	Total
Test Positive	51	14	65
Test Negative	10	2104	2114
Total	61	2118	2179

Overall % agreement = (51 + 2104)/(2179)*100%=98.9% (CI 98.4%–99.4%)

Positive % agreement = (51 / 61)*100% = 83.6% (CI 73.5%–93.7%)

Negative % agreement = (2104 / 2118)*100% = 99.3% (CI 99.0%–99.7%)

Range, mean and median values and lower percentiles for GSP Neonatal GALT kit and the Neonatal GALT kit (only data within measuring range of each device is included):

Method	N	Range	Mean	Median	0.5th percentile	1.0st percentile	1.5% percentile
Proposed (GSP) (U/dL)	2137	3.6-24.7	15.5	15.6	5.8	6.8	7.6
Predicate (U/g Hb)	2145	3.0-17.9	10.2	10.2	5.1	5.7	6.1

b. Matrix comparison:

Not applicable. This device only uses dried blood spots from newborns.

3. Clinical studies:

   a. Clinical Sensitivity:

   Not applicable.

   b. Clinical specificity:

   Not applicable

   c. Other clinical supportive data (when a. and b. are not applicable):

   Not applicable.

4. Clinical cut-off:

   Screening specimens that result in values at or below cut-off should be considered as presumptive positives for classical galactosemia and retested immediately. If upon retest the GALT value is still at or below cut-off, the specimen should be considered screen positive for classical galactosemia. Confirmation of screen positives should be by a diagnostic test procedure.

   If an assay is available to test for total galactose, it should also be run. If the specimen is positive for the total galactose assay, the specimen should be considered positive for classical galactosemia. Follow local requirements for follow-up testing.

5. Expected values/Reference range:

   GALT values by percentile from the testing completed with the Neonatal GALT kit at a U.S. state laboratory:

	n	Range	Mean	Median	Lower percentiles						
					5%	2.5%	2.0%	1.5%	1.0%	0.5%	0.25%
GSP Neonatal GALT (U/dL)	2137	3.6-24.7	15.5	15.6	9.7	8.6	8	7.6	6.8	5.8	5.2

   It should, however, be remembered that cut-off values of GALT in dry blood spots may vary between different tests and different populations. Therefore, it is recommended that each laboratory establishes its own reference range and cut-off limit from a representative sample population.

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

O. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.